Overexpression of Plant Specific Insert from cardosin B (PSI B) in Arabidopsis correlates with cell responses to stresses

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Abstract: Under abiotic stress, several changes occur in cells both regarding their physiology and cellular mechanisms. Plants have developed modifications in the production and trafficking of proteins and remodelling of endomembranes to overcome stress conditions. The alteration of the targeting of proteins to the vacuole by shifting their transport towards an unconventional, Golgi-independent, route is a good example. Plant Specific Inserts (PSIs) are known to mediate such routes and our goal was to evaluate if transgenic Arabidopsis plants overexpressing PSI B respond differently when subjected to different abiotic stresses (osmotic, oxidative, salt, and metal). The results obtained point to a differential expression of PSI B-mCherry depending on the type of stress and a decrease of cellular and cytoplasmatic movement in all stress conditions.

Keywords: Abiotic stress; Plant Specific Insert; protein trafficking; unconventional routes.
Introduction

Climate change

Abiotic Stress

- Stomatal closure
- Decreased photosynthetic activity
- Altered cell wall elasticity
- ROS production
Introduction

Plant Specific Insert

Independent domain, with approximately 100 aminoacidic

Highly conserved in innumerous species

ability to interact with membranes

redirect secreted proteins to the vacuole through different pathways
Goal:

Study the expression levels of PSIB and its biosynthetic pathways in transgenic Arabidopsis plants overexpressing this domain coupled to m-Cherry fluorescent protein, in physiological and under abiotic stress conditions.
Experiments

Arabidopsis thaliana transformation

Germination in different stress conditions

Root biometrical analysis

cDNA Preparation

qPCR

Stress Conditions

- S1 - 50 mM NaCl
- S2 - 100 mM NaCl
- H1 – 50 mM Manitol
- H2 – 100 mM Manitol
- Ox - 0,5 mM H2O2
- Zn – 150 μM ZNSO4
Results

PSI B expression changes in plants under abiotic stress

- S1- decrease in root development in both plants (Wt and PSI B)
- S2- significative increase in the size of roots in plants overexpressing PSI B
- H1 and H2- same tendency as S2
- Ox and Zn- no changes are observed
Results

PSI B expression changes in plants under abiotic stress

- PSI B-mCherry is **upregulated** in both H1, H2 and S1
- PSI B-mCherry is **downregulated** in Ox and Zn
Discussion

PSI B expression changes in plants under abiotic stress

At high salt conditions, there is a significant increase in the root length in the transformed plants, suggesting an increased stress tolerance that may be triggered at certain concentrations.

The higher and mild water stress conditions also showed to have significant changes in roots length.

Increased tolerance in order to mitigate the negative effects of environmental adversities.
Discussion

PSI B expression changes in plants under abiotic stress

The downregulation of PSI B-mCherry on Ox and Zn stress, reinforces the importance of this domain in Water and salinity stress, and may indicate a role in processes related with cellular homeostasis and water control.

Overexpression in salt and mild hydric stress suggests that degradation of storage proteins occurs in order to overcome and tolerate these restricted conditions.

The PSI may then be associated with the defense and developmental system, and yet operate by distinct mechanisms.
Discussion

PSI B expression changes in plants under abiotic stress

Analysis of PSI B-mCherry localization in roots showed a marked decrease in intracellular and cytoplasmic movements under all stress conditions.

Can vesicle movement be inhibited by stress?
Discussion

PSI B expression changes in plants under abiotic stress
Conclusion

PSI B has an active role in adaptation and eventually tolerance mechanisms against abiotic stress
Supplementary Materials

Movement of PSI B-mCherry-labeled compartment

Control

H1
Supplementary Materials

Movement of PSI B-mCherry-labeled compartment
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